

DEVELOPMENT OF PURE AND ADULTERATED HONEY
MEASUREMENT SYSTEM USING MULTIPLE
FREQUENCIES SIX-PORT REFLECTOMETER (SPR)

SITI AISHAH BINTI BAHARUDDIN

UNIVERSITI MALAYSIA PERLIS

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**DEVELOPMENT OF PURE AND ADULTERATED
HONEY MEASUREMENT SYSTEM USING
MULTIPLE FREQUENCIES SIX-PORT
REFLECTOMETER (SPR)**

by

**SITI AISHAH BINTI BAHARUDDIN
(1430611551)**

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UNIVERSITI MALAYSIA PERLIS

DECLARATION OF THESIS

Author's full name : SITI AISHAH BINTI BAHARUDDIN
Date of birth : 11.08.1990
Title : DEVELOPMENT OF PURE AND ADULTERATED HONEY
MEASUREMENT SYSTEM USING MULTIPLE FREQUENCIES
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LIST OF ABBREVIATIONS

ADC	Analog-digital converter
EM	Electromagnetic
GHz	Gigahertz
GI	Glycemix Index
GUI	Graphical User Interface
HG	Honey Gold (<i>Madu Lebah Asli</i>)
MHz	Megahertz
MUT	Material under test
PNA	Performance Network Analyzer
SMA	SubMiniature version A
SPR	Six-Port Reflectometer
spp.	species
TH	Trigona Honey (<i>Madu Kelulut</i>)

LIST OF SYMBOLS

ϵ^*	Dielectric permittivity
ϵ'	Dielectric constant
ϵ''	Loss factor
Γ	Complex reflection coefficient
$ \Gamma $	Reflection coefficient in magnitude
ϕ	Reflection coefficient in phase
$^{\circ}\text{C}$	Degree Celcius
Ω	Resistance (Ohms)
μm	micrometer
mm	millimeter
cm	centimeter
$\tan \delta$	loss tangent
q_i	reflection coefficient at each detection port i
k_i	calibration constant at each detection port i
p_i	incident power at each detection port i
R^2	coefficient of determination

Pembangunan Sistem Pengukuran Madu Asli dan Madu Adulterasi Menggunakan Pelbagai Frekuensi Meter Pantulan Enam Pangkalan

ABSTRAK

Tesis ini membentangkan tentang pembangunan Meter Pantulan Enam Pangkalan dalam mengklasifikasikan madu asli dan madu adulterasi melalui ukuran pantulan. Adulterasi madu adalah satu isu yang membimbangkan pengguna dari segi kesihatan, oleh itu adalah amat penting untuk mengawal kualiti madu. Salah satu cara untuk mengklasifikasikan madu adalah dengan teknik pantulan gelombang mikro kerana ia adalah mudah, pantas dan tidak merosakkan bahan. Namun, disebabkan PNA Penganalisa Rangkaian adalah besar dan mahal, oleh itu, Meter Pantulan Enam Pangkalan diperkenalkan. Dalam kerja ini, pengurangan kos dan saiz amat dititikberatkan dalam rekabentuk litar lima pangkalan. Litar mikrostrip Lima Pangkalan direkabentuk menggunakan perisian AWR Microwave Office 2002. Litar yang direka turut berfungsi bersama penukaran analog ke digital, pengesan sepaksi hujung terbuka, pengesan diod dan komputer untuk dijadikan sebagai Meter Pantulan Enam Pangkalan. Ukuran pantulan dijalankan ke atas Madu Lebah Asli dan Madu Kelulut menggunakan Meter Pantulan Enam Pangkalan dalam pelbagai frekuensi iaitu 0.60 GHz, 0.64 GHz, 0.80 GHz, 2.28 GHz, 2.42 GHz, 2.82 GHz, 3.47 GHz, 3.75 GHz and 4.21 GHz kerana kebanyakan SPR hanya berfungsi pada satu frekuensi. Prestasi litar dalam ukuran pantulan disahkan oleh Agilent E8362B PNA Penganalisa Rangkaian. Ukuran dielektrik juga dilakukan untuk menyiasat perubahan dielektrik terhadap kandungan air dan sukrosa didalam madu. Ia boleh dikesan bahawa pemalar dielektrik, ϵ' menurun apabila frekuensi bertambah. Pada masa yang sama, ϵ'' menurun dengan pertambahan kandungan air atau sukrosa untuk Madu Lebah Asli dan Madu Kelulut. Sementara itu, pada adulterasi air pula, faktor kehilangan, ϵ'' menurun dengan pertambahan frekuensi. Tambahan pula, ϵ'' menurun apabila kandungan air adalah $< 36\%$ dan $< 43\%$ untuk Madu Lebah Asli dan Madu Kelulut. Ia juga boleh didapati bahawa pada 1 GHz hingga 4 GHz, ϵ'' menaik apabila kandungan sukrosa bertambah untuk Madu Lebah Asli dan Madu Kelulut. Dalam ukuran pantulan, nilai magnitud pantulan, $|\Gamma|$ menurun apabila frekuensi bertambah untuk semua peratus kandungan air atau sukrosa bagi kedua-dua jenis madu. Dari segi fasa, $-\phi$ meningkat apabila frekuensi bertambah untuk adulterasi air kedua-dua jenis madu. Bagi adulterasi sukrosa, tiada perubahan yang ketara pada $-\phi$ apabila kandungan sukrosa bertambah bagi kedua-dua jenis madu. Tambahan pula, prestasi litar yang direka adalah baik dari segi $|\Gamma|$ dan ϕ . Purata ralat mutlak dalam ukuran adulterasi air diantara PNA Penganalisa Rangkaian dan Meter Pantulan Enam Pangkalan untuk kedua-dua madu adalah kurang daripada 0.15 pada $|\Gamma|$ dan 17° pada ϕ . Manakala, purata ralat mutlak dalam ukuran adulterasi sukrosa diantara PNA Penganalisa Rangkaian dan Meter Pantulan Enam Pangkalan untuk kedua-dua jenis madu adalah kurang daripada 0.13 pada $|\Gamma|$ dan 14° pada ϕ . Hal ini dapat membuktikan bahawa Meter Pantulan Enam Pangkalan adalah salah satu alternatif yang dapat menggantikan PNA Penganalisa Rangkaian yang memerlukan ruang yang besar dan mahal. Meter Pantulan Enam Pangkalan adalah ringkas, cepat, tepat dan merupakan sistem instrumentasi mudah alih dalam mengklasifikasikan antara madu asli dan madu adulterasi.

Development of Pure and Adulterated Honey Measurement System using Multiple Frequencies Six-Port Reflectometer (SPR)

ABSTRACT

This thesis presents a study on the development of multiple frequencies Six-Port Reflectometer (SPR) in characterizing pure and adulterated honey through reflection measurement. As honey adulteration is one of major health concern among consumer, thus it has paramount importance in inspecting the quality of honey. One of proposed methods is to characterize honey by using microwave reflection technique because this technique is simple, rapid and non-destructive. However, due to bulky and expensive PNA network analyzer, thus, SPR is introduced. In this work, reduction of cost and compactness are the main concern in designing Five-Port ring junction circuit. The microstrip Five-Port ring junction circuit was designed using AWR Microwave Office 2002. The fabricated circuit works with PICO analogue-digital converter (ADC), open-ended coaxial sensor, diode detectors and computing machine to form a complete SPR measurement system. The reflection measurements were conducted on Honey Gold and Trigona Honey for multiple frequencies of 0.60 GHz, 0.64 GHz, 0.80 GHz, 2.28 GHz, 2.42 GHz, 2.82 GHz, 3.47 GHz, 3.75 GHz and 4.21 GHz since most of the developed SPR implement at single frequency. The performance of SPR in reflection measurement was verified by Agilent E8362B PNA Network Analyzer. Besides, dielectric measurement was conducted to investigate its dielectric behavior due to water and sucrose content in honey. It can be noticed that dielectric constant, ϵ' decreases when frequency increases. In the meantime, ϵ' decreases with increases water and sucrose content for Honey Gold and Trigona Honey. Meanwhile, for water adulterated Honey Gold and Trigona Honey, loss factor, ϵ'' decrease when frequencies increases. In addition, ϵ'' decreases when water content $< 36\%$ and $< 43\%$ for Honey Gold and Trigona Honey, respectively. It can be found that at 1 GHz to 4 GHz, ϵ'' increases when sucrose content increases which applicable for Honey Gold and Trigona Honey. In reflection measurement, magnitude of reflection coefficient, $|\Gamma|$ decrease when frequency increases for all percentage of water and sucrose content for both honeys. Withal, phase, $-\phi$ increases as frequency increases for both water adulterated honeys. $-\phi$ varies insignificantly when sucrose content increases for both sucrose adulterated honeys. Furthermore, it was found that all fabricated circuits have good agreement in measurement for $|\Gamma|$ and ϕ . The measurement accuracy of water adulterated between PNA and SPR for both honeys exhibit absolute error less than 0.15 in $|\Gamma|$ and absolute error of 17° in ϕ . Meanwhile, sucrose adulterated honey measurement provides the high accuracy in measurement of sucrose content with absolute error for $|\Gamma|$ is less than 0.13 and in ϕ is 14° . On the other hand, all fabricated SPR circuits are able to provide an accurate prediction in terms of water content and sucrose content. This study shows that the SPR can be an alternative in replacing the bulky and expensive PNA. The SPR is a simple, fast, accurate and portable instrumentation system for characterize pure and adulterated honey.

CHAPTER 1

INTRODUCTION

1.1 Research Background

Honey is sweet, viscous and rich of nutrients produced by bees from nectar of plants or honeydew (Anklam, 1998). Honey is also highly prized for its powerful medicinal and health giving properties. It is also has been used for treatment of burn wounds and ulcers (Vandamme, Heyneman, Hoeksema, Verbelen, & Monstrey, 2013). However, there is one major issue in honey industry which is honey adulteration. This is because pure honey which provides anti-inflammatory and antifungal properties is very expensive (Moniruzzaman, Sulaiman, Khalil, & Gan, 2013).

Conventionally, honey quality is measured by human sensory method which mainly detects its color, viscosity, smell, and flavor (Diacu & Tantaveanu, 2007). However, the human sensory method is not scientific as it is dependent on subjective judgement which based on experience of an individual. On the other hand, chemical analyses, e.g. chromatography (Ruiz-Matute, Rodríguez-Sánchez, Sanz, & Martínez-Castro, 2010), high performance liquid chromatography (Tahsin et al., 2014) and mass spectrometry (Cabañero, Recio, & Rupérez, 2006) are used to characterize the honey, but it requires high performance and expensive instruments, long processing time and complicated procedures. In other words, chemical analyses limit the process of measurement in lab scale.

In recent years, microwave technique is widely known in characterizing material such as food and agricultural products. This microwave technique is a non-destructive, simple

and fast processing technique. Dielectric properties of food materials can be considered as major contributing factors to understand the interaction between microwaves and food products (Ahmed, Prabhu, Raghavan, & Ngadi, 2007). Dielectric properties of food materials reflect the storage and dissipation of EM energy (Icier & Baysal, 2004). There are several food products have been investigated dielectrically, e.g. sea cucumber (Cong, Liu, Tang, & Xue, 2012), corn flour (Bansal, Dhaliwal, & Mann, 2015), and peanut kernels (Zhang, Zhou, Ling, & Wang, 2016). Dielectric properties in food material are usually influenced by moisture, frequency, temperature, density and the physical state of food (Galema, 1997). These dielectric properties had given great achievement in food technology as well as agricultural industry by providing the knowledge related to the behaviour of food material. For instance, this dielectric properties information is very useful for pasteurization and characterization in honey (Guo, Zhu, Liu, & Zhuang, 2010).

Over past decades, commercial network analyzer is commonly used to characterize any materials at microwave frequencies through the measurement of S-parameter (Kissinger, Laemmle, Nasr, & Weigel, 2013). However, this network analyzer is bulky, expensive, not suitable for the in-situ measurement and has complex embedded architecture (Lee, Abbas, Nur Shahrizan, & Cheng, 2010). As the microwave reflection technique is one of the most promising direct techniques in food technology and agriculture, therefore it is possible to design a reliable, cost-effective in-situ measurement system to characterize between the pure and adulterated honey.

1.2 Problem Statement

Honey adulteration is very crucial as it degrades the nutrients in honey which is harmful to consumer who fully reliant on honey product for their health purpose (Socha, Galkowska, Bugaj, & Juszczak, 2015). Thus, it is essential to inspect the quality of honey.

Due to the bulky and expensive network analyzer, Six-Port Reflectometer (SPR) has been introduced as an alternative low cost solution (Engen, 1977) to the industry need of network analyzer. Initially, SPR's circuit consists of six-ports, however in recent years, six-ports has been reduced to five-ports by using Five-Port ring junction circuit due to the signal source redundancy (Riblet & Hansson, 1981) and it can meet the portability requirement. Unfortunately, in many literatures, SPR's circuit was designed at a particular frequency (Julrat, et al, 2012) which it limits the feasibility of circuit in other applications. Hence, Five-Port ring junction circuits which can work with multiple frequencies are designed and explored in this work.

Water content and sucrose content in pure and adulterated honey are hard to be distinguished and identified through its appearance. However, they can be identified through reflection measurement using SPR due to the variation of dielectric properties. The variation is caused by presence of water and sucrose content in honeys which is highly associated with dielectric properties. Meanwhile, complex reflection coefficient, Γ is function of dielectric constant, ϵ' and loss factor, ϵ'' (Lee, Chung, You, Cheng, & Zulkifly, 2014). As a result, water and sucrose content in adulterated honeys can be determined through reflection measurement in terms of magnitude ($|\Gamma|$) and phase (ϕ) of reflection coefficient, respectively. Therefore, in this work, Six-Port Reflectometer (SPR) with multiple frequencies is developed to measure the complex reflection coefficient, Γ of honey samples through the reflection technique measurement.

1.3 Objectives

- i. To design and develop a Five-Port ring junction circuit in multiple frequencies use.
- ii. To develop a microwave measurement system to characterize pure and adulterated honey.
- iii. To characterize pure and adulterated honey in terms of water content and sucrose content through dielectric properties and complex reflection coefficient, Γ .

1.4 Scope of Research

Layout of Five-Port ring junction circuit was designed in different dimensions by using AWR Microwave Office 2002 for the required specification to fulfill Riblet & Hansson requirement. The performance of S-parameters was investigated in different frequencies to ensure it comply with the Riblet & Hansson theory. The Five-Port ring junction circuit was fabricated. The fabricated Five-Port ring junction circuit was verified using Agilent E8362B PNA Microwave Network Analyzer. The performance of S-Parameter of fabricated circuits at multiple frequencies were identified by comparing to the S-Parameter in simulation.

Five-Port ring junction circuit was then assembled with PICO Analogue-Digital Converter (ADC), three Keysight 8471E diode detectors, and low loss coaxial cables to be SPR. Meanwhile, Agilent VEE Pro 6.0 was used as a platform for interface between SPR and computing machine. Furthermore, MATLAB server has been implemented in Agilent VEE Pro 6.0 to conduct six-port algorithm. Performance of developed SPR was compared with Agilent E8362B PNA Microwave Network Analyzer through measurement of known loads, i.e. air, water, methanol and ethanol.

Pure Honey Gold and Pure Trigona Honey were adulterated with water and sucrose as water and sucrose adulterated honeys. These prepared samples were measured in terms of magnitude, $|\Gamma|$ and phase, ϕ of reflection coefficient using SPR at multiple frequencies i.e. 0.60 GHz, 0.64 GHz, 0.80 GHz, 2.28 GHz, 2.42 GHz, 2.82 GHz, 3.47 GHz, 3.75 GHz and 4.21 GHz at room temperature. The measured reading were taken for three times. These measured results were compared with PNA to verify its accuracy and efficiency of SPR in reflection measurement. In addition, dielectric properties were determined through PNA by measuring dielectric constant, ϵ' and loss factor, ϵ'' . The dielectric properties of the prepared samples were measured at 1 GHz to 5 GHz at room temperature. The measured reading were taken for three times. The relationship among dielectric properties, reflection coefficient, sucrose content and water content of pure and adulterated honeys were established mathematically. However, chemical analysis and study on environment effects have not been considered in this work.

1.5 Outline of Thesis

Chapter 1 provides a brief introduction on background, problems and the aim of research work. The scope of the research project are discussed as well.

Chapter 2 explores the fundamental of Six-Port technique and ring junction circuit. This chapter also provides a brief elaboration on honey. The literatures were review on past works, existing and current solutions in identifying authenticity of honeys.

Chapter 3 of this thesis elucidates about the work flow of experimental work in this research, e.g. ring junction circuit design, ring junction circuit fabrication, sample preparation, dielectric and reflection measurement. The measurement technique and its procedures were described here. The performance of Five-Port junction circuit was also

verified through comparison between simulation and theoretical values in term of S-parameter.

In Chapter 4, SPR is used to measure air, water, methanol and ethanol for verification of performance. SPR is verified through comparison between measured reflection coefficient using SPR and PNA. Subsequently, comparison between measured reflection coefficient of SPR and PNA was conducted on pure and adulterated honeys. Their behaviours of reflection coefficient are analysed through dielectric response. Lastly, mathematical expressions were developed to establish relationship of sucrose and water content in adulterated honeys with reflection coefficient.

Chapter 5 concludes and highlights about the performance of Five-Port ring junction circuits and theirs frequencies in relating water and sucrose content with reflection coefficient. The accuracy of SPR is summarized.

1.6 Summary

In this chapter, the main issue occurred in honey industry that is honey adulteration are discussed. Several techniques to inspect the quality of honey has been pointed out as well. Furthermore, this chapter highlights the aim and scope of this research.

In the next chapter, explanation on honey, fundamental of Six-Port technique, previous work, existing and current solution in characterizing pure and adulterated honey will be further discussed for better understanding on this research.

CHAPTER 2

LITERATURE REVIEW

2.1 Honey

2.1.1 Honey Adulteration

Due to the demand in honeys, some unethical food industry runner manipulated the contents in honey for maximum profits growth which leads to massive lost in business among honey harvester and hoax among consumers. There are three classes of commercial honey which are pure honey, adulterated honey and artificial honey (Herzfeld, 2015). Pure honey is made from raw honey while adulterated honey contains some pure honey with other ingredients. Artificial honey is made from sugar or corn syrups with additives and food coloring. The main compositions in honey are fructose, glucose followed by water and sucrose (Wang, 2007). Sucrose content in honey is low in cost compared to fructose and glucose (Guo, Liu, Zhu, & Wang, 2011a). Hence, sucrose has been common additive in honey adulteration. Sucrose which produced from sugar beet (Bogdanov & Martin, 2002; Guler, Bakan, Nisbet, & Yavuz, 2007; Elflein & Raezke, 2008; Guler et al., 2014) is applied. The process of honey adulteration can be done by directly adding the sucrose syrups into the honey (Ruiz-Matute, Soria, Martinez-Castro, & Sanz, 2007).

There are several techniques to inspect the quality in honey. Conventionally, sensory analysis is used to detect the quality of honey. This technique detects honey color intensity (Peng, Ling, Aniza, Wei, & Suan, 2014), flavor, smell, viscosity and crystallization (Molan, 1996; Singhal, Kulkarni, & Rege, 1997). However, this technique

has its disadvantages which are unreliable, required an expertise and not practical. According to Alvarez-Suarez, Tulipani, Romandini, Vidal, & Battino (2009), it mentioned that color plays important characteristic in controlling the quality of honey which measured by using conventional analytical techniques. Unfortunately, this technique is limited by high implementation expense and long processing time. Chemistry methods were also proposed widely in inspecting the honey quality i.e. high performance liquid chromatography (Tahsin et al., 2014; Wang et al., 2015), gas chromatography and mass spectrometry (Ruiz-Matute et al., 2010). Similarly, these methods required expensive experimental set up and chemical, and involve complicated procedures as well.

Recently, a microwave technique has widely known in food industry. Although there are many physical techniques such as acoustic and optical properties were implemented in characterizing food products, this microwave technique is preferable because it is simple, rapid, sensitive and non-destructive (Lizhi, Toyoda & Ihara, 2008). As a result, many researchers studied the dielectric properties of food and agricultural products. For instance, fat content in beef has been characterized by using microwave techniques (Ng et al., 2007; Zhao, Downey, & Donnell, 2016) for quality control in food processing. Furthermore, other food products such as fresh apple (Guo, Nelson, Trabelsi, & Kays, 2007), grape marc (Sólyom, Kraus, Mato, Gaukel, Schuchmann, 2013) and vegetables oil (Corach, Sorichetti, & Romano, 2013) have been investigated in terms of their dielectric properties as well. Meanwhile, the dielectric properties of honey have been studied as reported in (Guo et al., 2010).

2.1.2 Honey Bees: Apis Spp. and Stingless Spp.

Honey from honeybees is usually produced by the bee species genus i.e. *Apis* spp. The subspecies of *Apis* spp i.e. *Apis mellifera* is widely found in Europe and Asia (Guerrini et al., 2009) which commonly used for Apiculture.

Stingless bees are group belongs to five different genera viz. *Melipona*, *Trigona*, *Meliponula*, *Dectylurina* and *Lestrimelitta*. *Trigona* is one of the largest genus of stingless bees living in tropical (Michener, 2007). There are eleven subgenera of *Trigona* bee species and it has been estimated that the number of known stingless bees species in world is around 400 to 500. In Malaysia, it has been identified that 32 species of stingless bee are recorded in Malaysia (Michener, 2013). *Heterotrigonaitama* and *Geniotrigonathoracica* are the most *Trigona* bee species used for Meliponinculture in Malaysia (Kelly, Farisya, Kumara, & Marcela, 2014).

Honey from *Apis mellifera* and *Trigona* spp. are different in terms of color, viscosity and taste. *Trigona* spp. honey is less viscous and darker in color compared to *Apis mellifera* honey. This is because it contains higher levels of phenolic content than *Apis mellifera* honey (Peng et al., 2014). However, information on physicochemical and therapeutic properties of *Trigona* spp. are scarced (Rao, Krishnan, Salleh, & Gan, 2016).

2.1.3 Composition in Honey

The composition in honey may be differs in various honey because it depends on its floral source, origin and climatic conditions of region (Escuredo, Dobre, Fernández-González, & Seijo, 2014). Honey contains sugars, organic acids, various amino acids and biological components (Sadat, 2013). Honey consists of many compounds which primarily contain high concentration of complex mixture of sugars (Hafiz et al., 2012). The major component in complex mixture of sugars is fructose, glucose and sucrose. It has been stated that fructose and glucose content is not less than 60g/100g (60% in mass